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(54) Title: ANTICANCER AGENTS BASED ON REGULATION OF PROTEIN PRENYLATION

(57) Abstract: Oncoproteins such as Ras and RhoB are known to induce cell division in an unrestrained manner when such proteins are localized at the inner surface of a cancer cell membrane. The localization is effected by the prenylation reaction, whereby a hydrophobic group (e.g. farnesyl protein transferase). Deactivation of the prenylation enzyme through covalent modification can therefore ultimately result in the mitigation and/or cessation of cancer cell growth. Various prenylation inhibitors having the necessary structural groups to bond covalently, or essentially irreversibly, to the prenylation enzyme include carbonyl or thiocarbonyl compounds (or masked versions of these compounds) and alpha oxo-epoxides bonded to a hydrophobic, substrate-mimicking group. The carbonyl or thiocarbonyl compounds also contain a nucleofugal atom or group to enhance the tendency to form covalent bonds.

ANTICANCER AGENTS BASED ON REGULATION OF PROTEIN PRENYLATION

CROSS-REFERENCE TO RELATED APPLICATIONS

[0001] This application claims priority from Provisional Application Serial No. 60/241,955 filed October 23, 2000, the disclosure of which is hereby incorporated by reference in its entirety.

FIELD OF THE INVENTION

[0002] The present invention relates to the prevention and/or treatment of cancer by inhibiting enzyme-catalyzed prenylation reactions that allow localization of Ras, RhoB, and other proteins at the inner surfaces of cancer cell membranes and other intracellular locations, thereby causing unrestrained cell division.

BACKGROUND OF THE INVENTION

[0003] Approximately 25% of all human cancers result from a mutant gene that encodes a mutant form of the protein known as Ras. In cancer cells, Ras activates the cells to divide in an unrestrained manner. To induce cell division, Ras must be localized at the inner surface of the cancer cell membrane. This membrane localization of Ras is effected by attachment of a hydrophobic group, typically the farnesyl group, but possibly the related geranylgeranyl group. In either case, the group becomes attached to Ras enzymatically in a process known as prenylation. Thus, interference with prenylation of Ras has the potential to prevent Ras localization at the inner surface of the cancer cell membrane, resulting in the cessation of unrestrained cell division and/or reversion of the cancer cell to a normal phenotype.

[0004] The enzyme that attaches the farnesyl group to Ras, RhoB, and other proteins to facilitate the proper localization of these proteins in the cell is farnesyl protein transferase, also known as protein farnesyltransferase (referred to here as FTase). The farnesyl group becomes attached to Ras, RhoB, and other proteins by reaction with farnesyl diphosphate, also known as farnesyl pyrophosphate (referred to here as FPP). In other words, FTase catalyzes the reaction illustrated below for the Ras protein, in which the protein becomes attached to the farnesyl group by displacement of pyrophosphate ($P_2O_7^{4-}$, referred to here as PP_i):

$$\text{Ras} + \text{FPP} \xrightarrow{\text{FTase}} \text{farnesyl-Ras} + PP_i$$

[0005] Thus, a key target in a strategy to retard cancer cell proliferation is the enzyme FTase. By regulating FTase activity, Ras farnesylation, RhoB farnesylation and geranylgeranylation, and the prenylation of other proteins can be controlled. This can alter the intracellular distribution of these proteins and in turn prevent cancer cells from proliferating. Because normal cells also require FTase activity, the optimal regulation of prenyltransferase activity must be determined empirically.

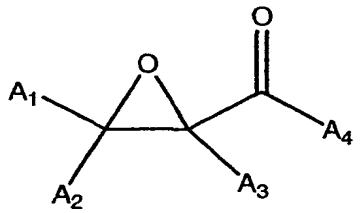
[0006] Many substances are known to block FTase activity and prevent farnesylation of cellular proteins. These include inhibitors of the enzyme FTase, which generally operate by blocking the binding of proteins to be prenylated, FPP, or both, to the FTase active site. Without the ability of the normal substrates (e.g. Ras and FPP) to bind to FTase, this enzyme can no longer transfer the farnesyl group from FPP to Ras. In general, inhibitors structurally mimic one or both of the natural substrates of the enzyme, in this case Ras and/or FPP. For conventional inhibitors, their binding to FTase is reversible and noncovalent (i.e. the binding of the inhibitor to FTase does not involve the formation of covalent bonds). Instead, hydrophobic forces, hydrogen

bonding, electrostatic attraction, etc. are principally responsible for binding of the inhibitor to the enzyme FTase. These binding forces allow the inhibitor to block the site on FTase where the normal substrates need to bind for farnesylation of Ras to occur.

[0007] It would therefore be desirable to develop a method of preventing, substantially irreversibly, FTase from farnesylating Ras and, more generally, preventing other prenylation enzymes from promoting the inner cell membrane localization of oncoproteins. Interaction of FTase, for example, with substances that covalently modify the active site of FTase should result in an enzyme with an essentially permanent reduction in catalytic ability. In principle, and in contrast to conventional enzyme deactivation, the covalent attachment can be irreversible or nearly irreversible. The desirable characteristics of a prenylation enzyme inhibitor may include both a substrate-mimicking group as well as a group having the ability to bond covalently to the enzyme at or near its active site.

SUMMARY OF THE INVENTION

[0008] The present invention is directed to methods for inhibiting a prenylation enzyme In one embodiment, the method comprises contacting a prenylation enzyme with a prenylation enzyme inhibitor of the following structural Formula I:

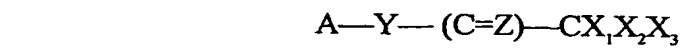


(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof,

where A_1, A_2, and A_3 are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, with the proviso that A_1, A_2, A_3, and A_4 are not all hydrogen. In a preferred embodiment, A_1, A_2, A_3, and A_4 are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl. In another preferred embodiment, at least one of A_1, A_2, A_3, and A_4 is a branched alkenyl, which may be specifically a farnesyl or geranylgeranyl group. In another preferred embodiment, the group A_4 is selected from the group consisting of hydrogen, alkyl (e.g. methyl), haloalkyl (e.g. trichloromethyl, trifluoromethyl, perfluoroethyl), aryl (e.g. phenyl), and heteroaryl (e.g. 4-pyridyl).

[0009] In another preferred embodiment, the prenylation enzyme inhibitor according to the method has the following structural Formula II:



(Formula II)

or a pharmaceutically acceptable salt, prodrug, or ester thereof,

where Y is a heteroatom or heteroatomic group selected from the group consisting of O, NH, NA', and S; where,

when Y is O, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA', A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

Z is oxygen or sulfur; and,

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, oxygen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen. In a preferred embodiment, A, and optionally A', are independently selected from the

group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl. In another preferred embodiment, at least one of A, and optionally A', is a branched alkenyl, which may be specifically a farnesyl or geranylgeranyl group. Preferably, at least one of X_1, X_2, and X_3 is a nucleofugal group capable of bonding with the active site of a prenylation enzyme.

[0010] According to the possibilities for Formula II, there exists a carbonyl variation, where Z is oxygen, and also a thiocarbonyl variation, where Z is sulfur. In the case of the carbonyl variation, a preferred class of prenylation inhibitors is obtained when X_1 is oxygen, such that the inhibitor will now have two carbonyl functions, and X_2 is methyl. These preferred compounds may also be categorized as pyruvic acid derivatives.

[0011] In another embodiment, a method of screening compounds as potential anti-tumor agents comprises contacting a prenylation enzyme with a test compound according to Formula I or Formula II. The method further comprises measuring prenylation activity of the enzyme to identify candidate anti-tumor agents. In a more specific embodiment, prior to the contacting step, a natural substrate of the prenylation enzyme (e.g. farnesyl pyrophosphate and/or geranylgeranyl pyrophosphate) is added to the test compound to compete therewith and indicate the specificity of the test compound for the prenylation enzyme.

[0012] In another embodiment, a method of inhibiting the growth of a cancer cell comprises contacting the cancer cell with a prenylation enzyme inhibitor according to Formula I or Formula II, where the growth of the cancer cell is inhibited.

[0013] In another embodiment, a pharmaceutically acceptable formulation comprises a compound according to Formula I or Formula II, and a pharmaceutically acceptable carrier.

[0014] These and other embodiments are described below in the detailed description of the invention.

DETAILED DESCRIPTION OF THE INVENTION

[0015] The present invention may hamper or prevent the proliferation of cancer cells, possibly resulting in a decrease in tumor size and/or disappearance of the cancer, to the benefit of cancer patients. It may act by interference with cancer cell biochemistry, in which the enzyme farnesyl protein transferase, geranylgeranyl protein transferase, and/or some other prenylation enzyme acts on the oncogenic Ras protein, RhoB protein, or some other growth-related cellular protein. Alteration of the ratio of farnesylated RhoB to geranylgeranylated RhoB, for instance, through the action of farnesyl protein transferase inhibitors, is thought to have a profound effect on cancer cell proliferation. The present invention may alter the farnesylation:geranylgeranylation ratio through the selective regulation of prenylation enzyme activity by covalent modification of the active site of prenylation enzymes rather than reversible inhibition of these enzymes, as is the current practice with farnesyl protein transferase reversible inhibitors.

[0016] The present invention is based on the effectiveness of prenylation enzyme inhibitors for ultimately reducing and/or terminating cancer cell proliferation through covalent or essentially irreversible modification of the enzyme. The potential advantages of the present invention over reversible inhibitors of prenyltransferase are: (1) buildup of unused substrate (e.g. FPP, Ras and RhoB proteins) cannot reverse the covalent bonding of this invention in the way reversible inhibitors can be displaced by unused substrates, making the present invention more effective; and (2) the effectiveness of periodic dosing with compounds of the present invention, in contrast to the need for the constant presence of reversible inhibitors, may (i) adjust the activity level of the enzyme, decreasing toxic side effects in the patient; and (ii) minimize the ability of the cancer cell to become resistant to the therapy.

[0017] The present invention can reduce or eliminate the unrestrained proliferation of cancer cells through the inhibition of enzymes affecting biochemical reactions in these cells. The specificity of the inhibitor for the target prenylation enzyme can be optimized through two parameters: (1) structural features, including structural similarity to the normal substrates (e.g. FPP and/or Ras), that direct or otherwise favor the binding of the inhibitor to the active site of enzyme, and (2) a reactivity that is appropriate for the chemical groups of the prenylation enzyme, particularly those that participate in the actual process catalyzed by the prenylation enzyme (e.g. those chemical groups that directly participate in the transfer of the farnesyl group of FPP to Ras). The structurally similar feature can be a hydrophobic component having a high affinity for the active site of the enzyme. For example, the active site of FTase is hydrophobic, which favors binding to its natural substrate FPP, also containing a hydrophobic region (i.e. the farnesyl group). Therefore, hydrophobic inhibitors that also contain an alpha-oxo epoxide or a carbon atom bearing at least one nucleofugal atom, where the carbon atom is bonded to a carbonyl (C=O) group or thiocarbonyl (C=S) group, or masked form of an alpha-oxo epoxide, carbonyl, or thiocarbonyl group, such as a group that will convert to a carbonyl or thiocarbonyl group at physiological conditions, are in many cases sufficiently reactive with the prenylation enzyme to inhibit its activity.

[0018] The term "alkyl," as used alone or in combination herein, refers to an unsubstituted or optionally substituted, straight, or branched chain saturated hydrocarbon group containing from one to twenty-five carbon atoms, preferably from one to fifteen carbons, such as methyl, ethyl, n-propyl, n-butyl, pentyl, hexyl, heptyl, octyl, the various branch chain isomers thereof, such as isopropyl, isobutyl, sec-butyl, tert-butyl, isohexyl and the like. The alkyl group may be optionally substituted by one or more substituents, and generally no more than three, and most often just one substituent. Preferred optional substituents include halo, alkoxy, amino, mono- and di-substituted amino, aryl, carboxylic acid, heterocyclo, heteroaryl, cycloalkyl, hydroxy, trifluoromethoxy and the like.

[0019] The term "lower alkyl," as used alone or in combination herein, refers to such alkyl groups containing from one to five carbon atoms.

[0020] The term "alkoxy," as used alone or in combination herein, refers to an alkyl group, as defined above, covalently bonded to the parent molecule through an -O- linkage, such as methoxy, ethoxy, propoxy, isopropoxy, butoxy, t-butoxy and the like.

[0021] The term "alkoxyalkyl," as used alone or in combination herein, refers specifically to an alkyl group substituted with an alkoxy group.

[0022] The term "aryloxy," as used alone or in combination herein, refers to an aryl group, as defined below, covalently bonded to the parent molecule through an -O- linkage. An example of an aryloxy is phenoxy.

[0023] The term "cycloalkoxy," as used alone or in combination herein, refers to a cycloalkyl group, as defined below, covalently bonded to the parent molecule through an -O- linkage.

[0024] The term "alkylthio," as used alone or in combination herein, refers to an alkyl group, as defined above, covalently bonded to the parent molecule through an -S- linkage.

[0025] The term "alkenyl," as used alone or in combination herein, refers to an alkyl group, as defined above, containing one or more carbon-carbon double bonds, preferably two or three double bonds. Examples of alkenyl include ethenyl, propenyl, 1, 3-butadienyl, and 1, 3, 5-hexatrienyl.

[0026] The term "alkynyl," as used alone or in combination herein, refers to an alkyl group, as defined above, containing one or more carbon-carbon triple bonds, preferably one or two such triple bonds.

[0027] The term "cycloalkyl," as used alone or in combination herein, refers to an unsubstituted or optionally substituted, saturated cyclic hydrocarbon group containing three to eight carbon atoms. The cycloalkyl group may optionally be substituted by one or more substituents, and generally no more than three, and most often just one

substituent. Preferred optional substituents include alkyl, halo, amino, mono- and di-substituted amino, aryl, hydroxy and the like.

[0028] The term "haloalkyl," as used alone or in combination herein, is a species of alkyl as defined herein, and particularly refers to an alkyl, preferably a lower alkyl, substituted with one or more halogen atoms, and preferably is a C_1 to C_4 alkyl substituted with one to three halogen atoms. One example of a haloalkyl is trifluoromethyl. Preferred examples of haloalkyl groups include trichloromethyl, triflouromethyl, and perflouroethyl.

[0029] The term "alkanoyl," as used alone or in combination herein, refers to an acyl radical derived from an alkanecarboxylic acid (alkyl-C(O)-), particularly a lower alkanecarboxylic acid, and includes such examples as acetyl, propionyl, butyryl, valeryl, and 4-methylvaleryl.

[0030] The term "aroyl," as used alone or in combination herein, means an acyl radical derived from an aromatic carboxylic acid, such as optionally substituted benzoic or naphthoic acids and specifically including benzoyl and 1-naphthoyl.

[0031] The term "aminocarbonyl," as used alone or in combination herein means an amino-substituted carbonyl (carbamoyl or carboxamide) wherein the amino group is a primary amino ($-NH_2$). Substituted aminocarbonyl refers to secondary (mono-substituted amino) or tertiary amino (disubstituted amino) group, as defined below, preferably having as a substituent(s) a lower alkyl group.

[0032] The term "aminoalkanoyl," as used alone or in combination herein, means an amino-substituted alkanoyl wherein the amino group is a primary amino group (-alkyl-C(O)-NH_2). The term "substituted aminoalkanoyl" refers to related secondary (mono-substituted amino) or tertiary amino (di-substituted amino) group, as defined below.

[0033] The term "carbocycloalkyl," as used alone or in combination herein, refers to an unsubstituted or optionally substituted, stable, saturated or partially unsaturated

monocyclic, bridged monocyclic, bicyclic, or spiro ring carbocycle of 3 to 15 carbon atoms such as cyclopropyl, cyclobutyl, cyclopentyl, cyclohexyl, cycloheptyl, bicyclohexyl, bicyclooctyl, bicyclononyl, spirononyl and spirodecyl. Cycloalkyls are thus one specific subset of carbocycloalkyls. The term "optionally substituted" as it refers to "carbocycloalkyl" herein indicates that the carbocycloalkyl group may be substituted at one or more substitutable ring positions by one or more groups independently selected from alkyl (preferably lower alkyl), alkoxy (preferably lower alkoxy), nitro, monoalkylamino (preferably a lower alkylamino), dialkylamino (preferably a di[lower]alkylamino), cyano, halo, haloalkyl (preferably trifluoromethyl), alkanoyl, aminocarbonyl, monoalkylaminocarbonyl, dialkylaminocarbonyl, alkylamido, (preferably a lower alkylamido), alkoxyalkyl (preferably a lower alkoxy[lower]alkyl), alkoxycarbonyl (preferably a lower alkoxycarbonyl), alkylcarbonyloxy (preferably a lower alkylcarbonyloxy) and aryl (preferably phenyl), said aryl being optionally substituted by halo, lower alkyl and/or lower alkoxy groups. Generally, there is no more than one optional substituent.

[0034] The term "heterocyclo," as used alone or in combination herein, refers to an unsubstituted or optionally substituted, stable, saturated, or partially unsaturated, monocyclic, bridged monocyclic, bicyclic, or spiro ring system containing carbon atoms and other atoms selected from nitrogen, sulfur and/or oxygen. Preferably, a heterocyclo group is a 5 or 6-membered monocyclic ring or an 8-11 membered bicyclic ring that consists of carbon atoms and contains one, two, or three heteroatoms selected from nitrogen, oxygen and/or sulfur. Heterocyclo includes bent-fused monocyclic cycloalkyl groups having at least one such heteroatom. The term "optionally substituted," as it refers to "heterocyclo" herein, indicates that the heterocyclo group may be substituted at one or more substitutable ring positions by one or more groups independently selected from alkyl (preferably lower alkyl and including haloalkyl (preferably trifluoromethyl)), alkoxy (preferably lower alkoxy), nitro, monoalkylamino (preferably a lower alkylamino), dialkylamino (preferably a di[lower]alkylamino), cyano, halo, alkanoyl, aminocarbonyl,

monoalkylaminocarbonyl, dialkylaminocarbonyl, alkylamido (preferably lower alkylamido), alkoxyalkyl (preferably a lower alkoxy[lower]alkyl), alkoxycarbonyl (preferably a lower alkoxycarbonyl), alkylcarbonyloxy (preferably a lower alkylcarbonyloxy) and aryl (preferably phenyl), said aryl being optionally substituted by halo, lower alkyl and lower alkoxy groups. Generally, there is no more than one optional substituent. The heterocyclo group may be, and generally is, attached to the parent structure through a carbon atom, or alternatively may be attached through any heteroatom of the heterocyclo group that results in a stable structure.

[0035] The term "heteroaryl," as used alone or in combination herein, refers to an unsubstituted or optionally substituted, stable, aromatic monocyclic or bicyclic ring system containing carbon atoms and other atoms selected from nitrogen, sulfur and/or oxygen. Preferably, a heteroaryl group is a 5- or 6-membered monocyclic ring (optionally benzofused) or an 8-11 membered bicyclic ring that consists of carbon atoms and contains one, two, or three heteroatoms selected from nitrogen, oxygen and/or sulfur. The term "optionally substituted" as it refers to "heteroaryl" herein indicates that the heteroaryl group may be substituted at one or more substitutable ring positions by one or more groups independently selected from alkyl (preferably lower alkyl and including haloalkyl (preferably trifluoromethyl)), alkoxy (preferably lower alkoxy), nitro, monoalkylamino (preferably a lower alkylamino), dialkylamino, (preferably a di[lower]alkylamino), cyano, halo, alkanoyl, aminocarbonyl, monoalkylaminocarbonyl, dialkylaminocarbonyl, alkylamido (preferably lower alkylamido), alkoxyalkyl (preferably a lower alkoxy[lower]alkyl), alkoxycarbonyl (preferably a lower alkoxycarbonyl), alkylcarbonyloxy (preferably a lower alkylcarbonyloxy), and aryl (preferably phenyl), said aryl being optionally substituted by halo, lower alkyl, and lower alkoxy groups. Generally, there is no more than one optional substituent. The heteroaryl group may be, and generally is attached to the parent structure through a carbon atom or alternatively may be attached through any heteroatom of the heteroaryl group that results in a stable structure. In the foregoing structures it is also contemplated that a nitrogen could be replaced with an N-oxide.

Both heterocyclo and heteroaryl also are intended to embrace benzo fused structures such as 1, 2-methylenedioxybenzene and 1, 4-benzodioxan. Preferred examples of heteroaryl groups include pyridyl (e.g. 2-, 3-, or 4-pyridyl).

[0036] The terms "halo" and "halogen," as used alone or in combination herein, represent fluorine, chlorine, bromine or iodine, preferably fluorine or chlorine for enzyme affinity, and preferably chlorine, bromine, or iodine when a nucleofuge.

[0037] The term "aryl," as used alone or in combination herein, refers to an unsubstituted or optionally substituted monocyclic or bicyclic aromatic hydrocarbon ring system having 6 to 12 ring carbon atoms. Preferred are optionally substituted phenyl, 1- naphthyl, or 2-naphthyl groups. The aryl group may optionally be substituted at one or more substitutable ring positions (generally at no more than three positions and most often at one or two positions) by one or more groups independently selected from alkyl (including haloalkyl (preferably trifluoromethyl and difluoromethyl)), alkenyl, alkynyl, alkoxy, aryloxy, nitro, hydroxy, amino, mono- and di-substituted amino, cyano, halo, alkanoyl, aminocarbonyl, carboxylic acid, carboxylic acid esters, carboxylic acid amide, an optionally substituted phenyl (optionally substituted by halo, lower alkyl and lower alkoxy groups), heterocyclo, or heteroaryl. Preferably, the aryl group is phenyl optionally substituted with up to four and more usually with one or two groups, preferably selected from lower alkyl, lower alkoxy, as well as cyano, trifluoromethyl, and halo.

[0038] The terms "aralkyl" and "(aryl)alkyl," as used alone or in combination herein, are species of alkyl as defined herein, and particularly refer to an alkyl group as defined above in which one hydrogen atom is replaced by an aryl group as defined above, and include benzyl, and 2-phenylethyl.

[0039] The terms "(heterocyclo)alkyl" and "(heteroaryl)alkyl," as used alone or in combination can be considered a species of alkyl as defined herein, and particularly refer to an to an alkyl group as defined above in which one hydrogen atom is replaced by a heterocyclo group as defined above, or by a heteroaryl group as defined above.

[0040] The terms "alkoxycarbonyl," as used alone or in combination herein, mean a radical of the formula -C(O)-alkoxy, in which alkoxy is as defined above.

[0041] The term "alkylcarbonyloxy," as used alone or in combination herein, means a radical of the formula -O-C(O)-alkyl, in which alkyl is as defined above.

[0042] The term "alkoxyalkanoyl," as used alone or in combination herein, means a radical of the formula -alkyl-C(O)-O-alkyl.

[0043] The term "carboxyalkyl," as used alone or in combination herein, means a radical of the formula --alkyl-C(O)-OH.

[0044] The term "substituted amino," as used alone or in combination herein, embraces both mono and di-substituted amino. These terms, alone, or in combination, mean a radical of the formula -NR'R", where, in the case of mono-substitution, one of R' and R" is a hydrogen and the other is selected from alkyl, cycloalkyl, aryl, heterocyclo, (aryl)alkyl, (heterocyclo)alkyl, heteroaryl and hetero(aryl)alkyl; in the case of di-substitution, R' and R" are independently selected from alkyl, cycloalkyl, aryl, heterocyclo, and heteroaryl, or R' and R" together with the nitrogen atom to which they are both attached form a three to eight-membered heterocyclo or heteroaryl radical.

[0045] The term "amido," as used alone or in combination herein, refers to the group (-NH-) and the term "substituted amido" embraces a radical of the formula (-NR'-) where R' has the meaning above in connection with substituted amino.

[0046] The terms "alkanoylamido," "aroylamido," "heterocyclocarbonylamido" and "heteroaroylamido," as used alone or in combination herein, mean groups of the formula R-C(O)-NH- where R is an alkyl, aryl, heteroaryl or heterocyclo group. The terms "heteroaroyl" and "heterocyclocarbonyl," when used alone or in combination, mean groups of the formula R-C(O)- where R is a heteroaryl or heterocyclo group.

[0047] Unless otherwise defined, the term "optionally substituted" as used herein, refers to the substitution of a ring system at one or more positions with one or more

groups selected from: C_1-C_5 alkyl, C_1-C_5 alkoxy, an optionally substituted phenyl, cyano, halo, trifluoromethyl, C_1-C_5 alkoxycarbonyl, C_1-C_5 alkyl carbonyloxy, mono- and bis-(C_1-C_5alkyl)-carboxamide, C_1-C_5 alkylamido, nitro, and mono- and bis-(C_1-C_5 alkyl) amino.

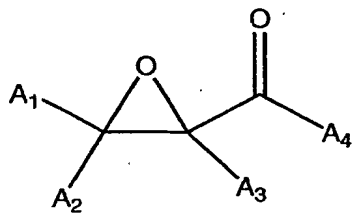
[0048] The terms "hydrophobic group" and "hydrophobic component" as used herein, refer to any of the groups hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido as defined above having at least some affinity for a hydrocarbon.

[0049] The terms "nucleofugal atom" and "nucleofugal group" as used herein refer to reactive leaving groups that, after reaction, can depart with a lone pair of electrons. Nucleofugal atoms or groups include halogen atoms (e.g. F, Cl, Br, and I) organosulfonyloxy groups (e.g. p-toluenesulfonyloxy, p-bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, etc.), acyloxy groups (e.g. CH_3CO_2—, CCl_3CO_2—), aryloxy groups (e.g. phenyl—O—), imidazolyl groups, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$.

[0050] It is recognized that there may be some overlap in some of the definitions of the various groups. Specific groups are mentioned, however, and may be particularly identified in the claims, in order to emphasize their positive inclusion in the described subject matter, as not only an optional substituent. As used herein, when a particular group, generally understood to have a single point of attachment to a core structure, such as an alkyl group, is identified in connection with a structure that must have two points of attachment in the structural core, it is understood that the named group (e.g.,

alkyl) refers to the parent group with a hydrogen or a site of unsaturation removed to create the second point of attachment so as to provide the required structure.

[0051] Compounds according to one embodiment of the present invention have the following generalized structural Formula I:



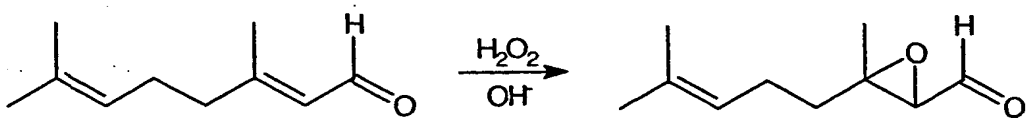
(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

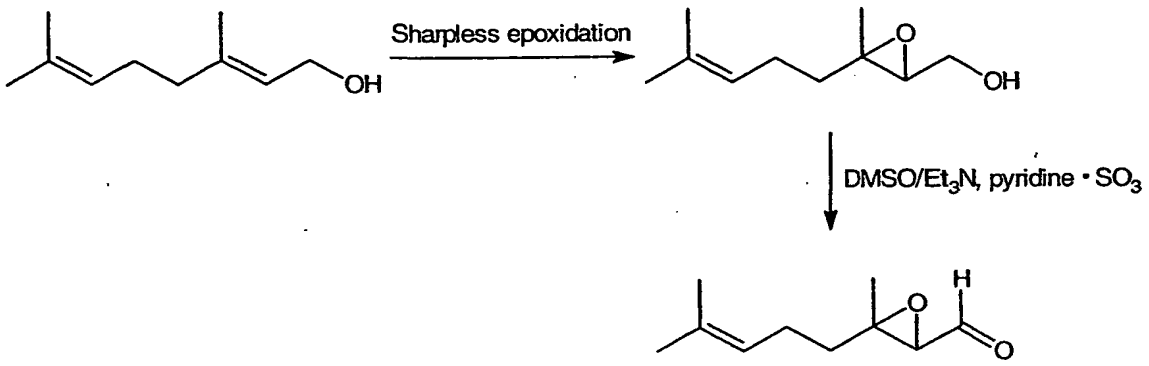
where A_1, A_2, and A_3, are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and A_1, A_2, A_3, and A_4 are not all hydrogen, since this would result in a compound lacking any specificity to the enzyme compared to the desired condition where at least one of these pendant groups has at least some specificity.

[0052] Preferably, at least one of A_1, A_2, A_3, and A_4 is a hydrophobic component designed to impart specificity of the substance for binding to and/or inactivation of FTase or GGTase. In this respect, in a preferred embodiment, A_1, A_2, A_3, and A_4 are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl. For example, at least one of A_1, A_2, A_3, and A_4 is a branched alkenyl, which might be specifically a farnesyl or geranylgeranyl group. In this case, the prenylation enzyme inhibitor has a pendant group that matches that of the substrates for which FTase and GGTase, respectively, have affinity. In another preferred embodiment, the group A_4 is selected from the group consisting of hydrogen, alkyl (e.g. methyl), haloalkyl (e.g. trichloromethyl, triflouromethyl, perflouroethyl), aryl (e.g. phenyl), and heteroaryl (e.g. 4-pyridyl). These particular groups for A_4 result in an inhibitor having improved ability to react with the active site of a prenylation enzyme. More specifically, and without being bound to any particular theory, the presence of halogen atoms in the group A_4 enhances the reactivity of the inhibitor with epsilon amino groups found in the prenylation enzyme active sites, and particularly the lysine epsilon amino group found in the active site of FTase. Additionally, the presence of phenyl and pyridyl groups enhance the stability of a Schiff base or imine that is believed to result from reaction of the inhibitor with the enzyme. From the above explanation, a particularly preferred variant of the prenylation enzyme inhibitor of the present invention according to Formula I is one in which at least one of A_1, A_2, and A_3 is a hydrophobic group (e.g. a farnesyl or a geranylgeranyl group) and A_4 is selected from the group consisting of alkyl, haloalkyl, aryl, and heteroaryl.

[0053] Possible methods of synthesizing prenylation inhibitor compounds of the present invention are provided below for compounds according to Formula I and Formula II as described above. In the case of alpha-oxo epoxides within the scope of Formula I, the treatment of the alpha,beta-unsaturated carbonyl compound with alkaline hydrogen peroxide, is depicted below:



[0054] Another synthesis possibility for compounds of Formula I is the Sharpless epoxidation of the allylic alcohol, followed by oxidation of the alcohol. This method results in the stereospecific formation of chiral alpha-oxo epoxides, and is depicted below:



[0055] Another class of compounds contemplated within the scope of the invention has the following generalized structural Formula II:

$$A—Y—(C=Z)—CX_1X_2X_3$$

(Formula II)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where Y is a heteroatom or heteroatomic group selected from the group consisting of O, NH, NA′, and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally

substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

Z is O or S, resulting in a carbonyl or thiocarbonyl group; and,

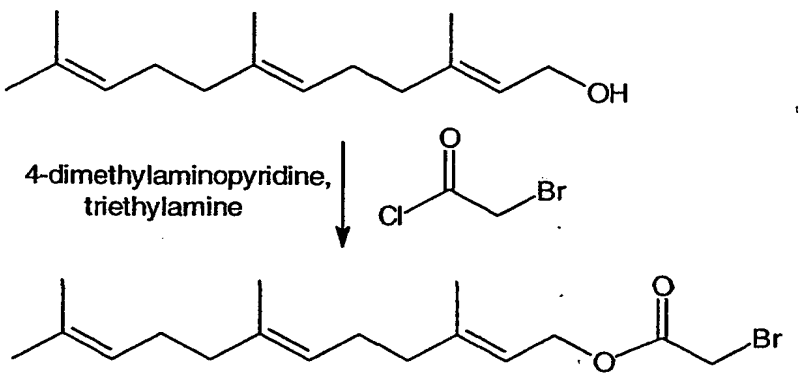
X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$. The groups X_1, X_2, and X_3 are not all hydrogen, as this particular case would result in the compound having an attached methyl group which is relatively unreactive compared to the desired condition where at least one of these pendant groups is a nucleofugal group. In a preferred embodiment, A, and optionally A′, are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl. In another preferred embodiment, at least one of A, and

optionally A′ is a branched alkenyl, which may be specifically a farnesyl or geranylgeranyl group.

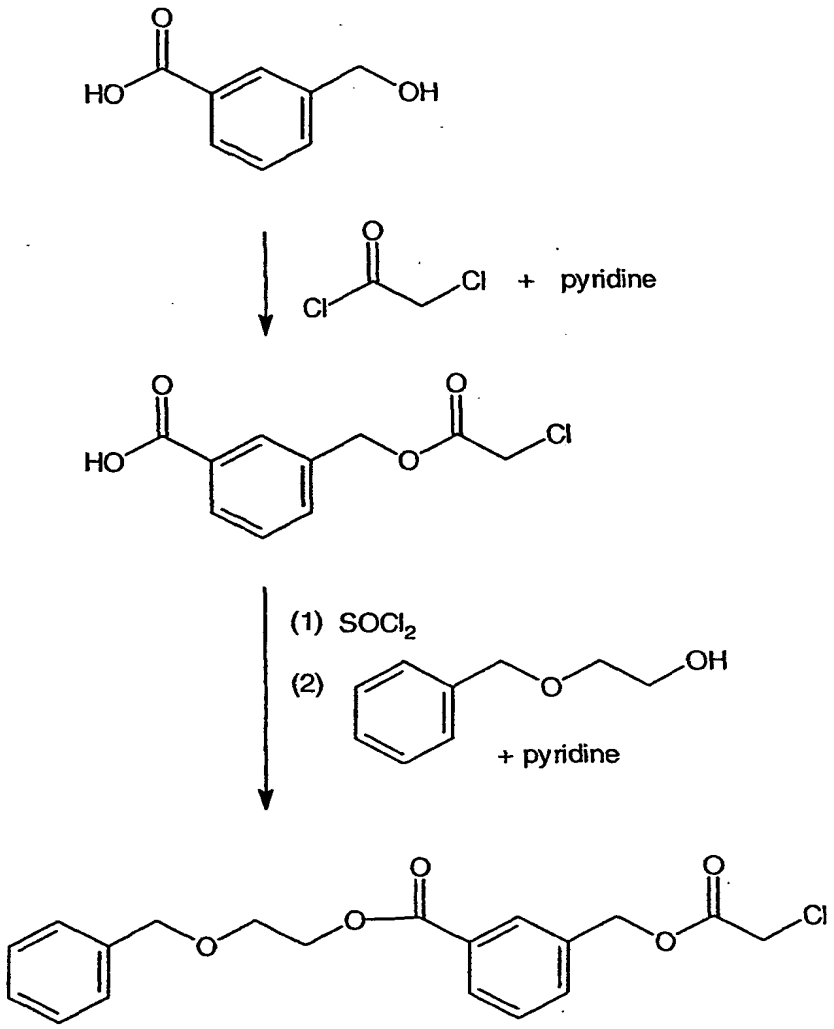
[0056] According to the possibilities for Formula II, there exists a carbonyl variation, where Z is oxygen, and also a thiocarbonyl variation, where Z is sulfur. Preferred enzyme inhibitor compounds within the generalized structural Formula II can be characterized as alpha-haloesters or alpha-halothioesters where Y is an oxygen atom, X_1 and X_2 are hydrogen, and X_3 is a halogen. In the case of the carbonyl variation of Formula II, a preferred class of prenylation inhibitors is obtained when X_1 is oxygen, such that the inhibitor will now have two carbonyl functions, and X_2 is methyl. These preferred compounds may also be categorized as pyruvic acid derivatives.

[0057] As mentioned, the portion of the compound represented by any of A aids in the selective, noncovalent binding or affinity of the inventive compounds for prenyltransferases. Also, the carbonyl or thiocarbonyl group of compounds represented by Formula II, combined with an adjacent nucleofuge-bearing carbon atom, can subsequently bond to the prenyltransferase active site, thereby regulating the level of activity of the prenyltransferase by hampering access of substrates to the active site residues and/or other mechanisms such as by inducing conformational changes in the prenyltransferase that affect its catalytic ability. The regulation of catalytic activity of the prenyltransferase can also be achieved by total inactivation of a portion of the prenyltransferase molecules while leaving some molecules completely unmodified.

[0058] It is possible to prepare compounds within the scope of Formula II according to the pathway provided below, where, for example, A is a farnesyl subsitutent, Y and Z are oxygen, X_1 and X_2 are hydrogen, and X_3 is bromine:



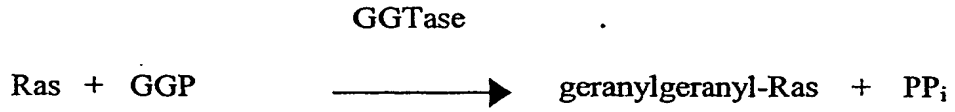
[0059] The pathway outlined below depicts the synthesis of another compound of Formula II, having a hydrophobic substituent comprising a phenyl group and a polyether functionality:



[0060] Other embodiments of the invention include a pharmaceutically acceptable salt, prodrug, or ester of the compounds of Formula I or Formula II. By way of example, for compounds having structural Formula II, the carbonyl groups can be masked in various forms including a hydrate [$C(OH)_2$], a hemiacetal or hemiketal [C(OH)(OR')], an acetal or a ketal [C(OR')(OR")], an acylal or related compound [C(OC(=O)R')OC(=O)R")], a bisulfite addition compound [$C(OH)(SO_3^-)$], an enol (C=COH), an enol ether (C=COR'), an enol ester [C=COC=(=O)R'], and so forth, wherein R' and R" are independently selected from the group consisting of hydrogen,

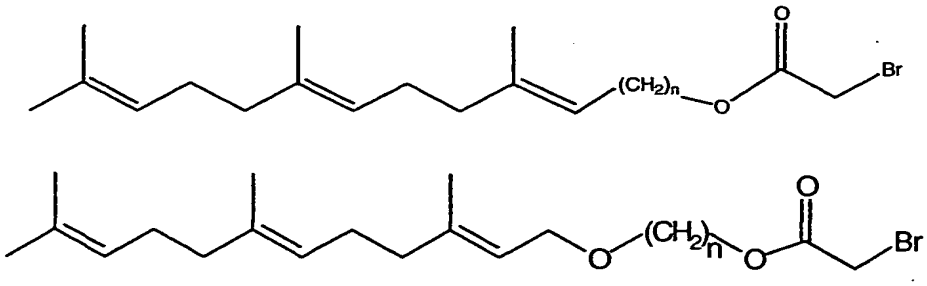
alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido. Likewise, prenylation enzyme inhibitors according to the thiocarbonyl variation of Formula II may also include masked thiocarbonyl groups, in which the oxygen atoms in the masked groups according to the above description are replaced by sulfur atoms. Of course, it is possible that, where the masked groups according to the above description contain more than one oxygen atom, a combination of oxygen and sulfur atoms may actually be included in the masked group. Such masked carbonyl or thiocarbonyl groups may produce the carbonyl or thiocarbonyl groups of the prenylation enzyme inhibitors of the present invention under physiological conditions.

[0061] Some cancer cells in which farnesylation of Ras is blocked alternatively employ the related prenylation reaction geranylgeranylation to attach a hydrophobic group to Ras, accomplishing membrane localization and continued cancerous behavior of the cell. The enzyme that attaches the geranylgeranyl group to Ras protein to facilitate localization at the inner surface of the cancer cell membrane is geranylgeranyl protein transferase, also known as protein geranylgeranyltransferase (referred to here as GGTase). The geranylgeranyl group becomes attached to Ras by reaction with geranylgeranyl diphosphate, also known as geranylgeranyl pyrophosphate (referred to here as GGPP). Stated otherwise, GGTase catalyzes the following reaction, in which Ras becomes attached to geranylgeranyl group by displacement of pyrophosphate ($P_2O_7^{4-}$, referred to here as PP_i):

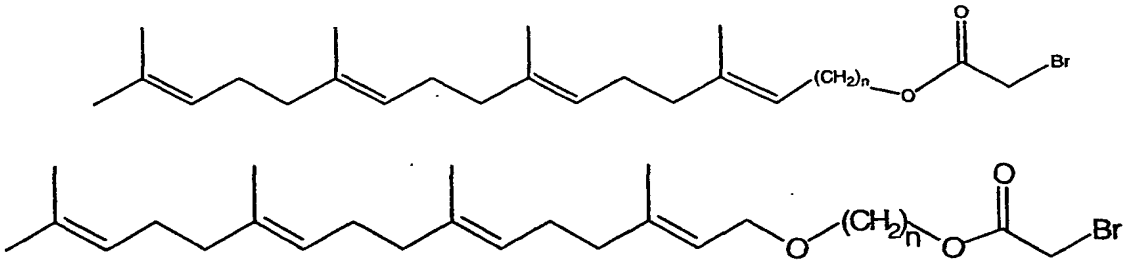


[0062] The newly formed geranylgeranyl-Ras localizes at the inner surface of the cancer cell membrane and causes the cancer cell to divide without restraint. Thus, a key target in a strategy to retard cancer cell proliferation is the enzyme GGTase. By reducing or destroying GGTase activity, either in combination with regulation of Ras farnesylation or independently, Ras geranylgeranylation may be also regulated, which in turn should further hinder the ability of the cancer cell to divide and proliferate through localization of Ras at its inner membrane surface. As noted previously, regulation of RhoB geranlygeranylation by use of inhibitors of the present invention is also contemplated as a means for retarding and/or terminating cancer cell growth.

[0063] To more precisely align the structure of the inhibitors of the present invention with the active sites of FTase, GGTase, or other enzymes, variation of the distance between the covalent-bonding group and the farnesyl-mimicking or geranylgeranyl-mimicking group is achieved through altering the length of a "spacer" between such groups. For example, inhibitors representative of the carbonyl variation of structural Formula II may be more precisely tailored to inhibit either FTase or GGTase activity by altering the spacer length as represented below:



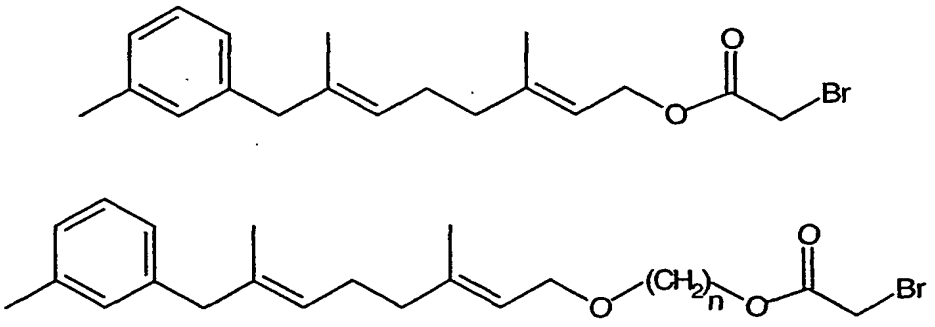
Targeted for FTase



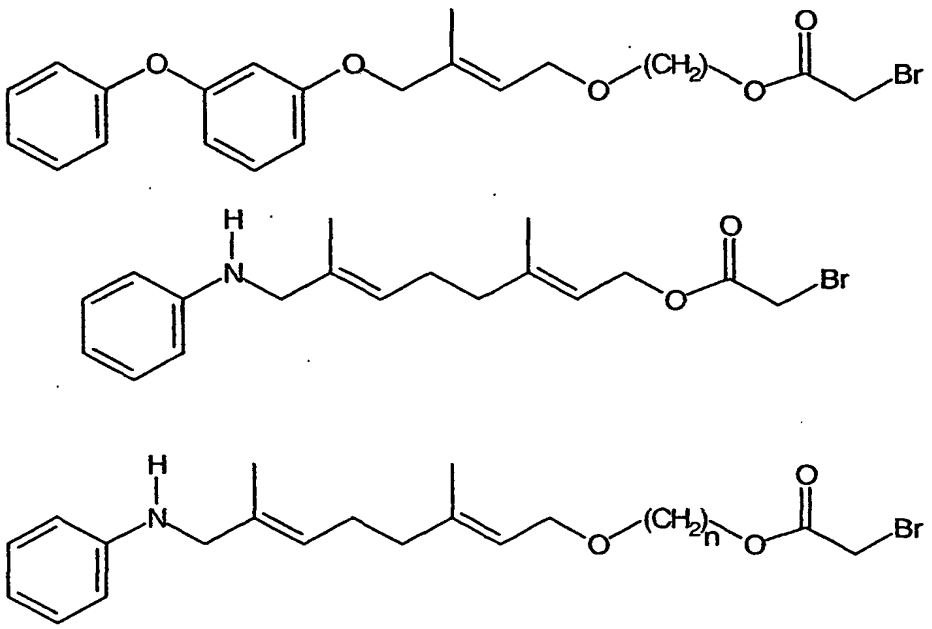
Targeted for GGTase

where the value of n, representing the number of carbon atoms between the respective farnesyl and geranylgeranyl groups, will generally range from 0 to about 10, although instances where longer chains are required for alignment with other types of enzymes are readily recognizable to the ordinary skilled artisan having regard for this disclosure.

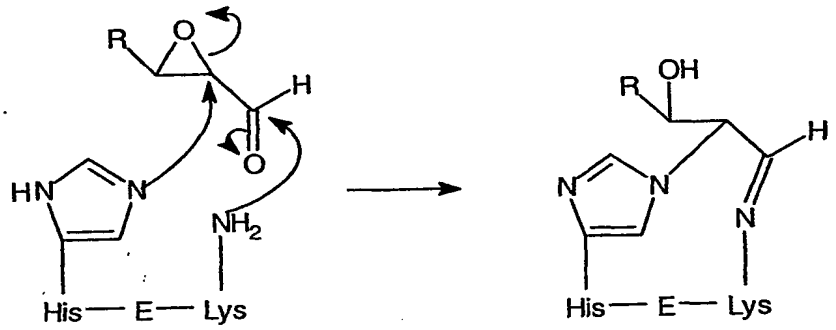
[0064] Inhibitors of the present invention may also incorporate an aromatic group for enhanced binding to the hydrophobic binding site of FTase or GGTase. Such compounds are exemplified by the following specific enzyme inhibitors according to the carbonyl variation of structural Formula II, although numerous other possible embodiments of this type of compound are of course possible and readily apparent to one of ordinary skill in the art, having regard for this disclosure:



[0065] Three additional possibilities are represented below, where the last two have an A group that is selective for the FTase active site over the GGTase and squalene synthase active sites, thus conferring added specificity for the desired target enzyme.



[0066] The inhibitors of the present invention are applicable in particular to the reduction in prenylation activity of the enzymes farnesyl protein transferase and geranylgeranyl protein transferase. Without wishing to be bound by any particular theory or reaction mechanism, a hypothetical pathway illustrating FTase inhibition using the alpha-oxo epoxide prenylation enzyme inhibitors, according to Formula I of the present invention, is shown below:



[0067] As shown, the FTase active site contains two amino acid residues that are involved in the reaction with the inhibitor, namely His-248 and Lys-294. The rest of the enzyme is conveniently represented as E. These two residues are in close proximity as a consequence of their normal role in binding the natural substrate FPP at its terminal phosphate. As shown above, nucleophilic attack by the His imidazole group results in epoxide ring opening, thus making the epoxide oxygen formally a nucleofuge. In addition to alkylation of the imidazole ring, the nearby epsilon-amino group of the Lys forms a Schiff base or imine with the aldehyde group of the alpha-oxo epoxide. The result of the above reactions is the crosslinking of two active site residues. The resulting crosslinked structure irreversibly precludes the FTase active site residues from catalyzing prenylation reactions, thus impairing the enzyme's overall functioning.

[0068] Analogous reaction mechanisms can be postulated for inhibitor compounds of the present invention according to both the carbonyl and thiocarbonyl variations of Formula II. In these cases, the nucleofugal atom or group adjacent to a carbonyl group can react with the aforementioned active site residues to crosslink them in an analogous manner to the mechanism shown above, with an accompanying release of the nucleofugal group.

[0069] Preferably, the inhibitor is administered under proper conditions and in a concentration such that its presence in the prenylation system reduces prenylation activity by at least about 50%, more preferably at least about 75%, and even more

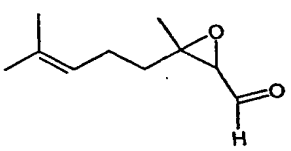
preferably by at least about 90%. Pharmaceutical formulations can be prepared by combining appropriate amounts of the inhibitor in a pharmaceutically acceptable carrier, diluent, or excipient. In such formulations, the inhibitor is typically present in an amount from about 0.1-20% by weight, and more commonly from about 1-10%.

[0070] Combinations of this invention with other anticancer agents to produce synergistic effects of benefit to the patient are also possible. This might be based on two strategies. One is to interfere with different biochemical processes to increase tumor cell killing. Another is to hamper development of drug resistance, which is less likely to occur simultaneously in tumor cells exposed to anticancer agents based on interference with different biochemical pathways in the tumor cells.

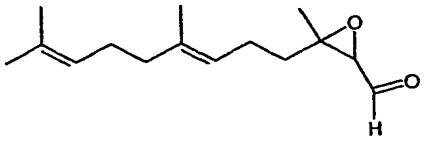
[0071] In summary an improved method of interference with protein prenylation in tumor cells has been described that may prevent or hamper the proliferation of tumor cells, possibly resulting in a decrease in tumor size and/or disappearance of the cancer, to the benefit of cancer patients.

EXAMPLES 1-2

[0072] Two alpho-oxo epoxides according to Formula I of the present invention were synthesized according to procedures set forth in H. Yao and D. E. Richardson, J. Am. Chem. Soc. 2000, 122, 3220-3221 and also in C. A. Bunton and G. J. Minkoff, J. Chem. Soc. 1949, 665-670. The compounds were then tested for their inhibition of the growth of various cancer cells. These compounds had, as their A_2 substituent, farnesyl and geranylgeranyl groups, respectively. These inhibitors had structural formulas as shown below.



EXAMPLE 1



EXAMPLE 2

[0073] The compounds were tested for growth inhibition in a variety of human cancer cell lines, according to procedures described by K.J. Okolotowicz, W.L. Lee, R.F. Hartman, S.R. Lefler, and S.D. Rose, *Inactivation of Protein Farnesyltransferase by Active-Site-Targeted Dicarbonyl Compounds*, Arch. Pharm. Pharm. Med. Chem. 2001, 334, 194-202. Results of biological testing of these compounds are summarized in Table 1, where the GI_{50} value represents the concentration of inhibitor compound required to effect a 50% reduction in cell growth (i.e. 50% inhibition).

Table 1

Inhibition of Cancer Cell Growth by Alpha-oxo Epoxides

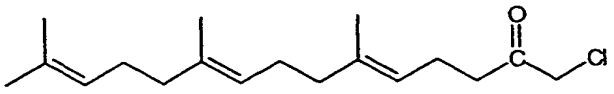
Human cancer cell line	GI_{50}/molar Compound RG-22	GI_{50}/molar Compound RG-23
Breast (MCF-7)	14×10^{-6}	8.9×10^{-6}
Prostate (DU-145)	27×10^{-6}	28×10^{-6}
Central nervous syst.(SF268)	30×10^{-6}	42×10^{-6}
Pancreatic (BXPC-3)	67×10^{-6}	51×10^{-6}
Colon (KM20L2)	95×10^{-6}	61×10^{-6}
Lung (NCI-H460)	117×10^{-6}	63×10^{-6}

[0074] Results show that both compounds are extremely active for inhibiting cell growth in various human cancer cell lines in culture. In fact, one of the GI_{50} values was below 10 micromoles/liter. Additionally, the compounds were active against mouse P388 leukemia cells in culture. In these cases, compound RG-22 exhibited an ED_{50} of 26×10^{-6} M, and RG-23 exhibited an ED_{50} of only 11×10^{-6} M. Based on these results, the anticancer activity of these compounds is confirmed.

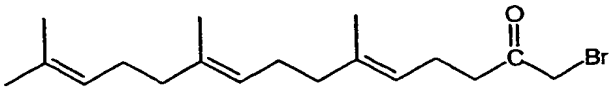
EXAMPLES 3-4

[0075] Two alpha-halo carbonyl compounds were also synthesized as decribed previously herein and tested for their inhibition of the growth of various cancer cells. These compounds had substituent farnesyl groups as well as substituent nucleofugal halogen atoms. The inhibitors had structural formulas as shown below:

EXAMPLE 3:



EXAMPLE 4:



[0076] The compounds were tested for growth inhibition in a variety of human cancer cell lines, as described in Examples 1-2. Results of the in vitro cell culture testing of these compounds are summarized in Table 2, which provides the associated GI_{50} values.

Table 2

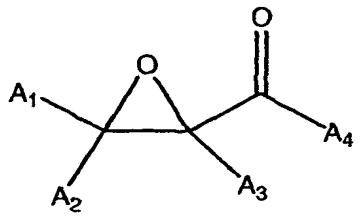
Cell Culture Results with Alpha-halo Carbonyl Compounds

Human cancer cell line	Example 3 GI_{50}/molar	Example 4 GI_{50}/molar
Colon (HT-29)	10^{-4} to 10^{-5}	10^{-4} to 10^{-5}
Colorectal (Colo-205)	10^{-4} to 10^{-5}	10^{-4} to 10^{-5}
NSC Lung (H-460)	1.1×10^{-5}	1.4×10^{-5}
Prostate (PC-3)	7.9×10^{-6}	1.6×10^{-5}
Acute myeloid leukemia (HL-60)	Above 10^{-4}	10^{-9} to 10^{-10}
Fibrosarcoma (HT-1080)	10^{-5} to 10^{-6}	10^{-4} to 10^{-5}
Urinary bladder (T-24)	10^{-4} to 10^{-5}	10^{-4} to 10^{-5}
Colon (Caco-2)	Above 10^{-4}	10^{-4} to 10^{-5}

[0077] Again, results show that the compounds provide effective inhibition of cancer cell growth, even at low concentrations.

WHAT IS CLAIMED IS:

1. A method for inhibiting a prenylation enzyme, the method comprising contacting the prenylation enzyme with a compound of Formula I:



(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where A_1, A_2, and A_3, are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and

A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl,

carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, with the proviso that A_1, A_2, A_3, and A_4 are not all hydrogen.

2. The method of claim 1 where the prenylation enzyme is farnesyl protein transferase or geranylgeranyl protein transferase.

3. The method of claim 1 where the activity of the prenylation enzyme is reduced by at least about 50%.

4. The method of claim 1 where the activity of the prenylation enzyme is reduced by at least about 75%.

5. The method of claim 1 where the activity of the prenylation enzyme is reduced by at least about 90%.

6. The method of claim 1 where A_4 is selected from the group consisting of hydrogen, alkyl, haloalkyl, aryl, and heteroaryl.

7. The method of claim 6 where A_4 is selected from the group consisting of methyl, trichloromethyl, triflouromethyl, perflouroethyl, phenyl, 2-pyridyl, 3-pyridyl, and 4-pyridyl.

8. The method of claim 1 where A_1, A_2, A_3, and A_4 are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl.

9. The method of claim 8 where at least one of A_1, A_2, A_3, and A_4 is a branched alkenyl group.

10. The method of claim 9 where at least one of A_1, A_2, A_3, and A_4 is farnesyl or geranylgeranyl.

11. A method for inhibiting a prenylation enzyme, the method comprising contacting the prenylation enzyme a compound of Formula IIA:

$$A—Y—(C=O)—CX_1X_2X_3 \quad \text{(Formula IIA)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA', and S; where,

when Y is O, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl,

carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA', A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen.

12. The method of claim 11 where the prenylation enzyme is farnesyl protein transferase or geranylgeranyl protein transferase.

13. The method of claim 11 where the activity of the prenylation enzyme is reduced by at least about 50%.

14. The method of claim 11 where the activity of the prenylation enzyme is reduced by at least about 75%.

15. The method of claim 11 where the activity of the prenylation enzyme is reduced by at least about 90%.

16. The method of claim 11 where Y is oxygen; X_1 and X_2 are hydrogen; and X_3 is a halogen.

17. The method of claim 11 where X_1 is O and X_2 is methyl.

18. The method of claim 11 where A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl.

19. The method of claim 18 where at least one of A and A′ is a branched alkenyl group

20. The method of claim 19 where at least one of A and A′ is farnesyl or geranylgeranyl.

21. A method for inhibiting a prenylation enzyme, the method comprising contacting the prenylation enzyme with a compound of Formula IIB:

$$A—Y—(C=S)—CX_1X_2X_3 \quad \text{(Formula IIB)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA′, and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen.

22. The method of claim 21 where the prenylation enzyme is farnesyl protein transferase or geranylgeranyl protein transferase.

23. The method of claim 21 where the activity of the prenylation enzyme is reduced by at least about 50%.

24. The method of claim 21 where the activity of the prenylation enzyme is reduced by at least about 75%.

25. The method of claim 21 where the activity of the prenylation enzyme is reduced by at least about 90%.

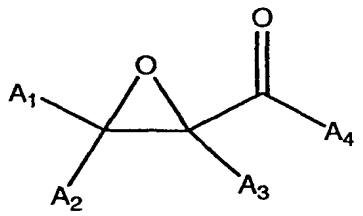
26. The method of claim 21 where Y is oxygen; X_1 and X_2 are hydrogen; and X_3 is a halogen.

27. The method of claim 21 where A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkenyl, alkynyl, aryl, aralkyl.

28. The method of claim 27 where at least one of A and A′ is a branched alkenyl group.

29. The method of claim 28 where at least one of A and A′ is farnesyl or geranylgeranyl.

30. A method of screening compounds as potential anti-tumor agents, the method comprising contacting a prenylation enzyme with a test compound of Formula I:



(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where A_1, A_2, and A_3, are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and

A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, with the proviso that A_1, A_2, A_3, and A_4 are not all hydrogen,

and measuring prenylation activity of the enzyme, where a reduction in prenylation activity renders the test compound a candidate anti-tumor agent.

31. The method of claim 30 where the reduction in prenylation activity is at least about 50% to render the test compound a candidate anti-tumor agent.

32. The method of claim 30 where a reduction in prenylation activity is at least about 75% to render the test compound a candidate anti-tumor agent.

33. The method of claim 30 where a reduction in prenylation activity is at least about 90% to render the test compound a candidate anti-tumor agent.

34. The method of claim 30 where the prenylation enzyme is farnesyl protein transferase geranylgeranyl protein transferase.

35. The method of claim 30 where, prior to the contacting step, a natural substrate of the prenylation enzyme is added to the test compound to compete therewith and indicate the specificity of the test compound for the prenylation enzyme.

36. The method of claim 35 where the natural substrate of the prenylation enzyme is farnesyl pyrophosphate or geranylgeranyl pyrophosphate.

37. A method of screening compounds as potential anti-tumor agents, the method comprising contacting a prenylation enzyme with a test compound of Formula IIA:

$$A—Y—(C=O)—CX_1X_2X_3 \quad \text{(Formula IIA)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA', and S; where,

when Y is O, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA', A and A' are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen,

and measuring prenylation activity of the enzyme, where a reduction in prenylation activity renders the test compound a candidate anti-tumor agent.

38. The method of claim 37 where the reduction in prenylation activity is at least about 50% to render the test compound a candidate anti-tumor agent.

39. The method of claim 37 where a reduction in prenylation activity is at least about 75% to render the test compound a candidate anti-tumor agent.

40. The method of claim 37 where a reduction in prenylation activity is at least about 90% to render the test compound a candidate anti-tumor agent.

41. The method of claim 37 where the prenylation enzyme is farnesyl protein transferase geranylgeranyl protein transferase.

42. The method of claim 37 where, prior to the contacting step, a natural substrate of the prenylation enzyme is added to the test compound to compete therewith and indicate the specificity of the test compound for the prenylation enzyme.

43. The method of claim 42 where the natural substrate of the prenylation enzyme is farnesyl pyrophosphate or geranylgeranyl pyrophosphate.

44. A method of screening compounds as potential anti-tumor agents, the method comprising contacting a prenylation enzyme with a test compound of Formula IIB:

$$A—Y—(C=S)—CX_1X_2X_3 \quad \text{(Formula IIB)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group selected from the group consisting of O, NH, NA′, and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio,

alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen,

and measuring prenylation activity of the enzyme, where a reduction in prenylation activity renders the test compound a candidate anti-tumor agent.or having the structural formula.

45. The method of claim 44 where the reduction in prenylation activity is at least about 50% to render the test compound a candidate anti-tumor agent.

46. The method of claim 44 where a reduction in prenylation activity is at least about 75% to render the test compound a candidate anti-tumor agent.

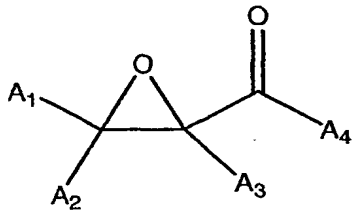
47. The method of claim 44 where a reduction in prenylation activity is at least about 90% to render the test compound a candidate anti-tumor agent.

48. The method of claim 44 where the prenylation enzyme is farnesyl protein transferase geranylgeranyl protein transferase.

49. The method of claim 44 where, prior to the contacting step, a natural substrate of the prenylation enzyme is added to the test compound to compete therewith and indicate the specificity of the test compound for the prenylation enzyme.

50. The method of claim 49 where the natural substrate of the prenylation enzyme is farnesyl pyrophosphate or geranylgeranyl pyrophosphate.

51. A method of inhibiting the growth of a cancer cell, the method comprising contacting the cancer cell with a compound of Formula I:



(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where A_1, A_2, and A_3, are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and

A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, with the proviso that A_1, A_2, A_3, and A_4 are not all hydrogen.

52. The method of claim 52 where the cancer cell is selected from the group consisting of colon, colorectal, NSC lung, prostate, acute myeloid leukemia, fibrosarcoma, and urinary bladder cancer cells.

53. The method of claim 52 where the GI_{50} concentration is less than about 10^{-4} molar.

54. A method of inhibiting the growth of a cancer cell, the method comprising contacting the cancer cell with a compound of Formula IIA:

$$A—Y—(C=O)—CX_1X_2X_3 \quad \text{(Formula IIA)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA′, and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally

substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen.

55. The method of claim 54 where the cancer cell is selected from the group consisting of colon, colorectal, NSC lung, prostate, acute myeloid leukemia, fibrosarcoma, and urinary bladder cancer cells.

56. The method of claim 54 where the GI_{50} concentration is less than about 10^{-4} molar.

57. A method of inhibiting the growth of a cancer cell, the method comprising contacting the cancer cell with a compound of Formula IIB:

$$A—Y—(C=S)—CX_1X_2X_3 \quad \text{(Formula IIB)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA′, and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio,

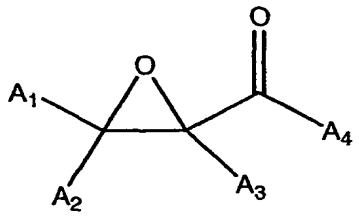
alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen.

58. The method of claim 57 where the cancer cell is selected from the group consisting of colon, colorectal, NSC lung, prostate, acute myeloid leukemia, fibrosarcoma, and urinary bladder cancer cells.

59. The method of claim 57 where the GI_{50} concentration is less than about 10^{-4} molar.

60. A pharmaceutical formulation comprising a compound of Formula I:



(Formula I)

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where A_1, A_2, and A_3, are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, and carboxyalkyl, and

A_4 is selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, with the proviso that A_1, A_2, A_3, and A_4 are not all hydrogen,

and a pharmaceutically acceptable carrier, diluent, or excipient.

61. A pharmaceutical formulation comprising a compound of Formula IIA:

$$A—Y—(C=O)—CX_1X_2X_3 \quad \text{(Formula IIA)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA', and S; where,

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen,

and a pharmaceutically acceptable carrier, diluent, or excipient.

62. A pharmaceutical formulation comprising a compound of Formula IIB:

$$A—Y—(C=S)—CX_1X_2X_3 \quad \text{(Formula IIB)}$$

or a pharmaceutically acceptable salt, prodrug, or ester thereof

where:

Y is a heteroatom or heteroatomic group independently selected from the group consisting of O, NH, NA′, and S; where

when Y is O, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxyalkyl, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido;

when Y is S, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, carbocycloalkyl or optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido; and,

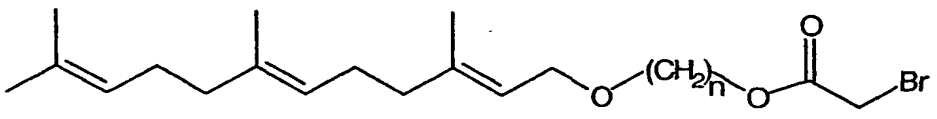
when Y is NH or NA′, A and A′ are independently selected from the group consisting of hydrogen, alkyl, alkoxy, alkoxyalkyl, aryloxy, cycloalkoxy, alkylthio, alkenyl, alkynyl, cycloalkyl, haloalkyl, alkanoyl, aroyl, aminocarbonyl, aminoalkanoyl or optionally substituted aminoalkanoyl, carbocycloalkyl or

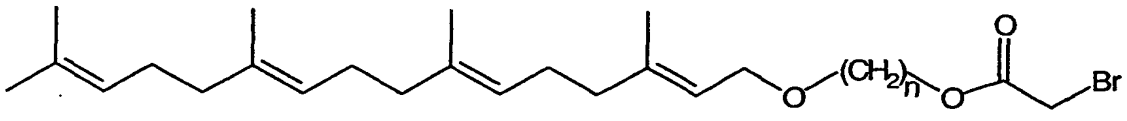
optionally substituted carbocycloalkyl, heterocyclo or optionally substituted heterocyclo, heteroaryl or optionally substituted heteroaryl, halo, aryl, aralkyl, (heterocyclo)alkyl, (heteroaryl)alkyl, alkoxycarbonyl, alkylcarbonyloxy, alkoxyalkanoyl, carboxyalkyl, amino or substituted amino, amido or substituted amido, and alkanoylamido, and

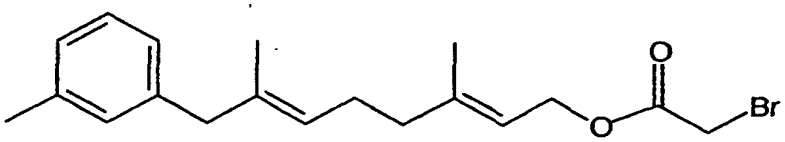
X_1, X_2, and X_3 are independently selected from the group consisting of hydrogen, halogen, organosulfonyloxy, bromobenzenesulfonyloxy, methanesulfonyloxy, trifluoromethanesulfonyloxy, acyloxy, aryloxy, imidazolyl, —O—N=O, —NO_2, —OSO_3^-, and —$OPO_2(OH)^-$, with the proviso that X_1, X_2, and X_3 are not all hydrogen,

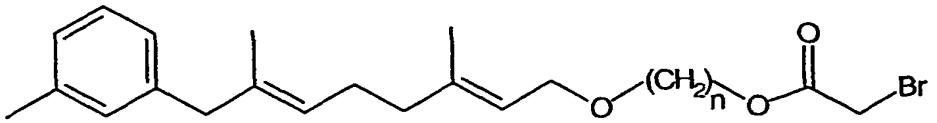
and a pharmaceutically acceptable carrier, diluent, or excipient.

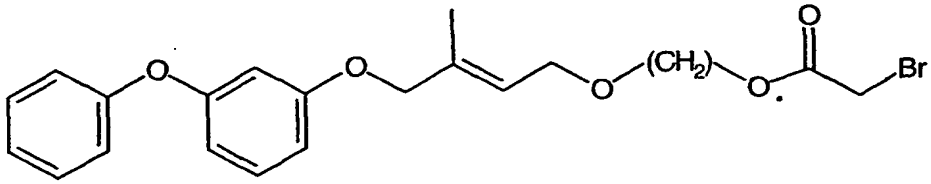
63. A compound having a formula selected from the group consisting of:

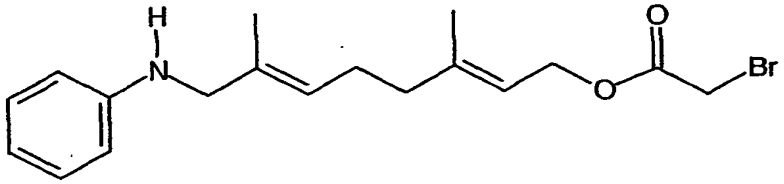




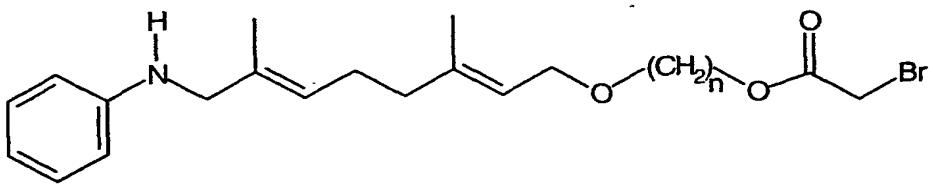








and



or a pharmaceutically acceptable salt, prodrug, or ester thereof.

64. A pharmaceutical composition comprising a compound of claim 63 and a pharmaceutically acceptable carrier, diluent, or excipient.

65. A method of inhibiting a prenylation enzyme comprising contacting the enzyme with a compound of claim 63.

66. A method of treating cancer comprising administering an effective amount of a pharmaceutical composition of claim 60.

67. A method of treating cancer comprising administering an effective amount of a pharmaceutical composition of claim 61.

68. A method of treating cancer comprising administering an effective amount of a pharmaceutical composition of claim 62.

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(81) Designated States *(national)*: AE, AG, AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, BZ, CA, CH, CN, CO, CR, CU, CZ, DE, DK, DM, DZ, EC, EE, ES, FI, GB, GD, GE, GH, GM, HR, HU, ID, IL, IN, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MA, MD, MG, MK, MN, MW, MX, MZ, NO, NZ, PH, PL, PT, RO, RU, SD, SE, SG, SI, SK, SL, TJ, TM, TR, TT, TZ, UA, UG, UZ, VN, YU, ZA, ZW.

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WO 02/034247 A3

(54) Title: ANTICANCER AGENTS BASED ON REGULATION OF PROTEIN PRENYLATION

(57) Abstract: Oncoproteins such as Ras and RhoB are known to induce cell division in an unrestrained manner when such proteins are localized at the inner surface of a cancer cell membrane. The localization is effected by the prenylation reaction, whereby a hydrophobic group (e.g. farnesyl protein transferase). Deactivation of the prenylation enzyme through covalent modification can therefore ultimately result in the mitigation and/or cessation of cancer cell growth. Various prenylation inhibitors having the necessary structural groups to bond covalently, or essentially irreversibly, to the prenylation enzyme include carbonyl or thiocarbonyl compounds (or masked versions of these compounds) and alpha oxo-epoxides bonded to a hydrophobic, substrate-mimicking group. The carbonyl or thiocarbonyl compounds also contain a nucleofugal atom or group to enhance the tendency to form covalent bonds.

INTERNATIONAL SEARCH REPORT

Inte il Application No
PCT/US 01/32618

A. CLASSIFICATION OF SUBJECT MATTER
IPC 7 C07D303/42 A61K31/00 C07C29/10

According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)
IPC 7 C07C C07D A61K

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

Electronic data base consulted during the international search (name of data base and, where practical, search terms used)
EPO-Internal, PAJ, WPI Data, BIOSIS, CHEM ABS Data

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	WO 00 04032 A (LAMOTHE MARIE ;PF MEDICAMENT (FR); HALAZY SERGE (FR)) 27 January 2000 (2000-01-27)	
A	WO 00 33826 A (HARTMAN ROSEMARIE F ;OTTERSBERG STEVEN R (US); ROSE SETH D (US); L) 15 June 2000 (2000-06-15)	
X	PATENT ABSTRACTS OF JAPAN vol. 1995, no. 04, 31 May 1995 (1995-05-31) & JP 07 002821 A (YAMANOUCHI PHARMACEUT CO LTD), 6 January 1995 (1995-01-06) abstract; example II	60

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[X] Further documents are listed in the continuation of box C.

[X] Patent family members are listed in annex.

° Special categories of cited documents :

"A" document defining the general state of the art which is not considered to be of particular relevance
"E" earlier document but published on or after the international filing date
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"X" document of particular relevance; the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance; the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art.
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
18 September 2002	30. 09. 02

Name and mailing address of the ISA	Authorized officer
European Patent Office, P.B. 5818 Patentlaan 2 NL – 2280 HV Rijswijk Tel. (+31–70) 340–2040, Tx. 31 651 epo nl, Fax: (+31–70) 340–3016	Deffner, C-A

Form PCT/ISA/210 (second sheet) (July 1992)

C.(Continuation) DOCUMENTS CONSIDERED TO BE RELEVANT

Category °	Citation of document, with indication,where appropriate, of the relevant passages	Relevant to claim No.
X	WO 93 22304 A (SMITHKLINE BEECHAM PLC ;GRIBBLE ANDREW DERRICK (GB); GROOT PIETER) 11 November 1993 (1993-11-11) page 11; example XVI	60
X	EP 0 248 597 A (ORTHO PHARMA CORP) 9 December 1987 (1987-12-09) page 25, line 34	60
X	EP 0 335 315 A (CIBA GEIGY AG) 4 October 1989 (1989-10-04) page 9; example XIIB page 17, line 48; example 19A	60
X	EP 0 410 244 A (BAYER AG) 30 January 1991 (1991-01-30) page 33; example 62	60
X	EP 0 147 217 A (MERCK FROSST CANADA INC) 3 July 1985 (1985-07-03) page 16, line 5	60
X	US 4 235 923 A (DURHAM HARRY G) 25 November 1980 (1980-11-25) claim 1	60
X	EP 0 170 048 A (CIBA GEIGY AG) 5 February 1986 (1986-02-05) page 60 -page 61; examples B2,TO,B5	60
X	US 1 599 532 A (HUMPHREYS CHARLES A) 14 September 1926 (1926-09-14) claim 1	60
X	WO 91 15473 A (FOURNIER SCA LAB ;CHAUVIN LAB SA (FR)) 17 October 1991 (1991-10-17) page 11; example VII	60
X	EP 0 534 546 A (MERCK & CO INC) 31 March 1993 (1993-03-31) claim 2	6
X	US 4 900 749 A (MATSUMOTO SAICHI ET AL) 13 February 1990 (1990-02-13) example 14; table 2	61
X	EP 0 752 465 A (PROCTER & GAMBLE) 8 January 1997 (1997-01-08) page 30 -page 43	61

INTERNATIONAL SEARCH REPORT

International application No.
PCT/US 01/32618

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This International Search Report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.:
because they relate to subject matter not required to be searched by this Authority, namely:

Although claims 1-10,51-53 and 11- 29, 54- 59, 65- 68 are directed to a method of treatment of the human/animal body, the search has been carried out and based on the alleged effects of the compound/composition.

2. [ ] Claims Nos.:
because they relate to parts of the International Application that do not comply with the prescribed requirements to such an extent that no meaningful International Search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

see additional sheet

1. [X] As all required additional search fees were timely paid by the applicant, this International Search Report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this International Search Report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this International Search Report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[X] No protest accompanied the payment of additional search fees.

FURTHER INFORMATION CONTINUED FROM PCT/ISA/ 210

This International Searching Authority found multiple (groups of) inventions in this international application, as follows:

1. Claims: 1-10,30-36,51-60

   Methods and formulations relating to formula I

2. Claims: 11-29,37-50,54-59,61-68

   Methods, compounds and formulations relating to formulae IIA and IIB

INTERNATIONAL SEARCH REPORT

Information on patent family members

Intern pplication No
PCT/US 01/32618

Patent document cited in search report		Publication date	Patent family member(s)		Publication date
WO 0004032	A	27-01-2000	FR	2781228 A1	21-01-2000
			AU	4788699 A	07-02-2000
			WO	0004032 A1	27-01-2000
WO 0033826	A	15-06-2000	AU	3113600 A	26-06-2000
			WO	0033826 A1	15-06-2000
JP 07002821	A	06-01-1995	NONE		
WO 9322304	A	11-11-1993	AU	4041293 A	29-11-1993
			CN	1092418 A	21-09-1994
			WO	9322304 A1	11-11-1993
			EP	0639187 A1	22-02-1995
			JP	8500085 T	09-01-1996
			SI	9300238 A	31-12-1993
			US	5447954 A	05-09-1995
EP 0248597	A	09-12-1987	DK	273687 A	30-11-1987
			EP	0248597 A2	09-12-1987
			JP	63002968 A	07-01-1988
			PT	84972 A ,B	01-06-1987
EP 0335315	A	04-10-1989	AT	107639 T	15-07-1994
			AU	3225289 A	12-10-1989
			CA	1340463 A1	23-03-1999
			CY	1967 A	04-07-1997
			DD	283617 A5	17-10-1990
			DE	58907920 D1	28-07-1994
			DK	148989 A	30-09-1989
			EP	0335315 A1	04-10-1989
			ES	2056137 T3	01-10-1994
			FI	891362 A ,B,	30-09-1989
			HK	192696 A	25-10-1996
			HR	940505 A1	28-02-1997
			HU	49599 A2	30-10-1989
			HU	9400046 A3	28-12-1994
			IE	66248 B1	27-12-1995
			JP	1299283 A	04-12-1989
			JP	2777183 B2	16-07-1998
			KR	125933 B1	26-12-1997
			MX	15429 A	28-02-1994
			NO	891286 A ,B,	02-10-1989
			NZ	228477 A	27-11-1990
			PT	90125 A ,B	10-11-1989
			SI	8910630 A ,B	30-04-1997
			US	5149717 A	22-09-1992
			YU	63089 A1	31-12-1990
			ZA	8902261 A	29-11-1989
EP 0410244	A	30-01-1991	CA	2021996 A1	28-01-1991
			DD	299173 A5	02-04-1992
			EP	0410244 A1	30-01-1991
			JP	3118320 A	20-05-1991
EP 0147217	A	03-07-1985	CA	1288107 A1	27-08-1991
			EP	0147217 A2	03-07-1985
			JP	60169432 A	02-09-1985
			US	4761425 A	02-08-1988

INTERNATIONAL SEARCH REPORT

Information on patent family members

Inter pplication No
PCT/US 01/32618

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
US 4235923 A	25-11-1980	AU	5791080 A	06-11-1980
		CA	1142523 A1	08-03-1983
		DK	191980 A	05-11-1980
		EP	0018682 A1	12-11-1980
		ZA	8002605 A	29-04-1981
EP 0170048 A	05-02-1986	AT	62668 T	15-05-1991
		AU	4422985 A	02-01-1986
		CA	1276393 A1	13-11-1990
		DE	3582527 D1	23-05-1991
		DK	291485 A	29-12-1985
		EP	0170048 A2	05-02-1986
		ES	544534 D0	16-04-1986
		ES	8606260 A1	01-10-1986
		FI	852527 A	29-12-1985
		GR	851563 A1	25-11-1985
		IL	75620 A	15-05-1989
		NO	852589 A ,B,	30-12-1985
		NZ	212573 A	29-03-1989
		PT	80716 A ,B	01-07-1985
		US	4649215 A	10-03-1987
		DD	243697 A5	11-03-1987
		HU	38304 A2	28-05-1986
		HU	196364 B	28-11-1988
		JP	61018756 A	27-01-1986
		ZA	8504856 A	26-02-1986
US 1599532 A	14-09-1926	NONE		
WO 9115473 A	17-10-1991	FR	2660655 A1	11-10-1991
		WO	9115473 A1	17-10-1991
EP 0534546 A	31-03-1993	US	5298655 A	29-03-1994
		CA	2079080 A1	28-03-1993
		EP	0534546 A1	31-03-1993
		JP	5271258 A	19-10-1993
		US	5362906 A	08-11-1994
US 4900749 A	13-02-1990	CA	1280414 A1	19-02-1991
		EP	0194901 A2	17-09-1986
		GB	2172286 A ,B	17-09-1986
		JP	62000052 A	06-01-1987
		KR	9102687 B1	03-05-1991
		US	4977170 A	11-12-1990
EP 0752465 A	08-01-1997	EP	0752465 A1	08-01-1997
		BR	9608747 A	17-02-1999
		CA	2222708 A1	05-12-1996
		CN	1192776 A	09-09-1998
		JP	11506486 T	08-06-1999
		WO	9638528 A1	05-12-1996